

## **Project Report Arizona Iceberg Lettuce Research Council**

**For period  
(July 2005 through June 2006)**

### **Breeding high quality and disease resistant iceberg lettuce cultivars for Arizona**

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Growers and shippers throughout the region

## **SUMMARY**

Our objectives are to incorporate disease resistance into crisphead lettuce cultivars and breeding lines adapted to low desert production conditions. Major breeding efforts targeted resistance to lettuce big vein, Fusarium root rot, lettuce drop (*Sclerotinia sclerotiorum*), tipburn, and powdery mildew. In 2005, we confirmed resistance of previously identified germplasm to big vein, lettuce drop, and powdery mildew; we obtained preliminary data on inheritance of resistance to Fusarium root rot. Advanced breeding lines were evaluated and selections were made for resistance to bolting, tipburn, big vein, powdery mildew, and lettuce drop.

## **INTRODUCTION**

Production of iceberg lettuce from December through April is centered in the low desert region of Arizona and California, and is critical to maintaining a year round supply of high quality lettuce. Lettuce production is continually faced with preexisting and new challenges; therefore, development of improved cultivars with superior adaptation and pest resistance is important for sustaining this industry. Our research emphasizes the development of landmark cultivars adapted to early, middle, or late season conditions with resistance to pests, physiological disorders and environmental stresses.

The USDA lettuce breeding project has an ongoing desert program, both in Arizona and in the Imperial Valley. Currently, the USDA is the only publicly funded lettuce breeding program

operating in the low desert. In recent years, most of the desert production has been in the Yuma area. Therefore nearly all of our on-site work has been done in Yuma and has focused on the production concerns of this region. The objectives have been to develop iceberg lettuce breeding lines possessing 1) bolting resistance for early fall production, 2) big vein, powdery mildew, and downy mildew resistance for mid-winter production, 3) tipburn resistance for late spring production, 4) lettuce drop resistance for fall, winter, and spring planting, and 5) Fusarium root rot resistance (Fusarium wilt) for fall, winter, and spring planting.

## **REPORTS ON SPECIFIC PROJECTS**

### **Big Vein Resistance**

Big vein is a serious viral disease in the low desert region, and the development of resistant cultivars is needed to reduce economic damage resulting from big vein. Partially resistant cultivars adapted for coastal California production environments have been released which have a reduced frequency of symptomatic plants and/or symptom expression that is delayed until plants reach market maturity. Partially resistant cultivars have not been developed for Yuma production conditions. Furthermore, progress in increasing the level of partial resistance through breeding has been slow. One important reason for this is a lack of genetic variation among resistant cultivars, which are all derived from a few USDA releases. New sources of resistance need to be identified so that higher levels of resistance can be developed.

Big vein is caused by Mirafiori Lettuce Big Vein Virus (MLBVV) which is vectored by the soil fungus *Olpidium brassicae*. Complete resistance or immunity to MLBVV has only been identified in accessions of *Lactuca virosa* L., a distant relative of lettuce. No cultivars possessing immunity from *L. virosa* have been developed. Within the USDA breeding program, *L. virosa* – *L. sativa* hybrid breeding lines have been identified that have a reduced percentage of symptomatic plants, a type of resistance similar to what is observed in cultivated lettuce. It is likely that the genes responsible for resistance in *L. virosa* – *L. sativa* hybrids are distinct from those in currently available cultivars. Introgression of this resistance into cultivated lettuce may lead to higher levels of resistance than what is currently available.

The objectives of big vein research are to:

- 1) Introgress partial resistance from cultivated lettuce into low desert adapted cultivars.
- 2) Identify new sources of resistance.
- 3) Introgress resistance from *L. virosa* into cultivated lettuce.

### **Field experiments**

#### **Procedures**

A mid-winter field trial was conducted in Yuma, AZ to identify breeding lines and cultivars with big vein resistance under mid-winter production conditions. The experiment was planted on

October 15; the incidence of big vein symptomatic plants was recorded on January 19 for five Winterhaven x Pacific breeding lines and 17 cultivars. Breeding lines with low incidence of big vein were retained for backcrossing and further selection. For comparison among the 19 cultivars, the mean percent symptomatic plants and 95% confidence intervals were calculated.

A field experiment was conducted in 2005 to evaluate resistance in materials derived from Pacific x (Pacific x Pavane). In 2004, 5  $F_{3:5}$  families were tested in Yuma, AZ and selections were made from all 5 families. In 2005, 19  $F_{5:6}$  breeding lines were tested in a big vein infested field experiment that was planted on October 15 and evaluated for big vein disease on January 19. The percentage of symptomatic plants was calculated for each line.

## Results and Discussion

Variation was observed among cultivars for the percentage of big vein symptomatic plants, although these differences were generally not significant (Table 1). Only the latin cultivar Pavane can be distinguished as significantly different from 7 other cultivars. Further testing will be conducted next season to confirm these results. Two out of the five Winterhaven x Pacific breeding lines had less than 30% symptomatic plants (data not shown). These two lines were backcrossed to low desert adapted breeding lines to improve type and for further selection of big vein resistance.

Two years of testing was completed for breeding lines derived from Pacific x (Pacific x Pavane). In 2004, 5  $F_{3:5}$  families were tested in Yuma, AZ. All of these lines were as or more resistant than Pacific, Winterselect, Winterhaven, or Vangaurd 75 (Table 2). In 2005, 19  $F_{5:6}$  lines were tested for resistance in a low desert field experiment. Fourteen lines had fewer symptomatic plants than Pacific, and three were more resistant than Winterselect (Table 2). Further testing of this material will be conducted to determine the potential to release this germplasm.

## Greenhouse experiments

### Procedures

The big vein resistance in *L. virosa* – *L. sativa* germplasm was investigated in greenhouse experiments. Two related lines, 03-366-3 and 03-366-9 were identified with a high level of resistance from 5 generations of selection. These lines along with Salinas 88, Pacific, Pavane, and Clemente, and *L. virosa* accession IVT280, were tested in a greenhouse experiment. In previous greenhouse experiments, 256  $BC_2F_3$  families from crosses between 03-366-3 or 03-366-9 by Salinas, Salinas 88, Pacific, Pavane, and Clemente were tested and 295 plants in 64 families were selected for resistance greater than Pavane. A greenhouse experiment was conducted to identify transgressive segregants (families that are more resistant than both parents) within populations of  $BC_2F_{3:4}$  *L. virosa* – *L. sativa* hybrid families. The existence of transgressive segregants indicates the presence of novel alleles from *L. virosa* are being introgressed into cultivated lettuce. Three replications of 64 families with 8 seedlings per line were tested.

New sources of big vein resistance are being identified. Ten plant introductions (PIs) that had demonstrated resistance in previous unreplicated greenhouse experiments were tested in a 2005 greenhouse experiment with 3 replications. The control cultivars Pavane, Pacific, Margarita (all resistant), and Great Lakes 65 (susceptible) were included.

Each greenhouse experiment was conducted by inoculating plants with MLBVV carrying *Olpidium brassicae* zoospores collected from roots of symptomatic lettuce plants. The inoculated seedlings are transplanted into infested soil and grown in a cool greenhouse. The incidence of symptomatic plants was recorded for up to 8 weeks, and used to calculate the percentage of symptomatic plants at the end of the experiment and the area under the disease progress curve. AUDPC data was analyzed using Proc Mixed in SAS and 95% confidence intervals were constructed to detect significant differences among treatments.

### Results and Discussion

Variation for AUDPC was found in a greenhouse test of 00-366-3, 00-366-9, *L. virosa* accession IVT280, and *L. sativa* controls (Table 3). As in previous experiments, IVT280 had no symptoms. Among partially resistant lines, Pacific, Pavane, and 00-366-3 were the most resistant. 00-366-9 had a higher AUDPC than 00-366-3, but the two lines could not be detected as significantly different. This result indicates that 00-366-3 and 00-366-9 are valuable sources of big vein resistance.

Sixty-four BC<sub>2</sub>F<sub>3,4</sub> families were tested for big vein resistance in a 2005 greenhouse experiment to identify transgressive segregates. Transgressive segregates were detected in 00-366-9 x Salinas and 00-366-9 x Salinas 88 (Table 4). Families from 00-366-3 x Salinas and 00-366-3 x Salinas 88 were found that were significantly different from Salinas or Salinas 88, but none were significantly better from the more resistant parent 00-366-3. Families derived from 00-366-3 x Pacific and Pavane and 00-366-9 x Clemente had AUDPCs ranging from 1.6 – 30.3, 0.8 – 23.4, 0.3 – 22.5 respectively. The low end of this variation includes families with substantially less disease than Pavane Pacific, or Clemente. However, the disease pressure does not appear to be high enough to distinguish these families as significantly different from their parents. Regardless, these data demonstrate the value of 00-366-3 and 00-366-9 in developing lettuce germplasm with improved big vein resistance, and indicate the high likelihood that *L. virosa* is contributing novel alleles for big vein resistance.

Ten PIs and 3 resistant cultivars varied for the percent symptomatic plants, and had significantly lower AUDPC than Great Lakes 65 (Table 5). Three PIs, PI342452c, 342555, and 324242 had sufficiently low levels of disease to warrant further testing.

### Powdery Mildew Resistance

Powdery mildew caused by the fungus *Erysiphe chioacearum* can damage lettuce production in mid-winter production conditions. Resistance can be an effective control of powdery mildew, although no resistant iceberg lettuce cultivars are known. Resistance is reported in butterhead and romaine cultivars, Imperial 850, and *L. serriola* PIs 234204 and 255665. Of these sources of

resistance, the inheritance of resistance is reported only for Imperial 850, which is conferred by a single dominant gene.

Our research objectives are to:

- 1) Introgress resistance from butterhead cultivars Big Boston, Bremex, Clarion, and Soraya into new cultivars and breeding lines.
- 2) Identify new sources of resistance for breeding.
- 3) Determine the genetic gain from greenhouse selection of powdery mildew resistance.

## **Identifying New Sources of Resistance and Breeding Line Development**

### **Procedures**

A field trial with three replications was planted and allowed to become naturally infested with *Erysiphe chicoacearum* to evaluate for powdery mildew resistance. Plots were evaluated on March 9 for powdery mildew severity using 1 through 5 scale (1 = no powder mildew – 5 = complete coverage of powdery mildew). Two experiments were conducted within this planting. These were to identify new sources of resistance in 49 leaf, romaine, iceberg, and primitive *Lactuca* germplasm and to select for powdery mildew resistance within 67 F<sub>3</sub> Salinas x Soraya, 7 F<sub>3</sub> Waldmann's Green x Soraya, and 4 F<sub>3</sub> Darkland x Clarion families.

### **Results and Discussion**

Moderate levels of powdery mildew infection were observed in the Yuma field trial. Variation for powdery mildew severity was observed among 49 leaf, romaine, iceberg, and primitive *Lactuca* germplasm. The 2005 experiment was a second years evaluation of modern germplasm for powdery mildew resistance. As in previous years, the most susceptible cultivars were icebergs, while the leaf types were rich sources of resistant cultivars. Several cultivars performed well in 2005 that were susceptible in 2004. These include Bremex, Imperial 850, and PI255665. Several cultivars performed well in both years, having powdery mildew severities less than 3. These cultivars include green leaf: Two Star, Waldmann's Green, Shining Star, Tehema, red leaf: Red Tide, Red Fox, butterhead: Clarion, Cindy, Soraya, Big Boston, Corelli and romaine: Parris Island and Green Towers.

Sixteen plants were selected from F<sub>3</sub> Salinas x Soraya for powdery mildew resistance. The resulting F<sub>4</sub> families will be retested the following season. No selections were made from Waldmann's green x Soraya or Darkland x Clarion crosses due to the availability of resistance in modern adapted romaine and leaf cultivars.

## **Genetic Gain from Greenhouse Selection of Powdery Mildew Resistance**

### **Procedures**

In previous experiments using a greenhouse test for powdery mildew severity, F<sub>2</sub> plants were selected as either resistant or susceptible. Three susceptible and eight resistant F<sub>3</sub> families derived from the greenhouse test were evaluated for powdery mildew severity in a naturally

infested field experiments with 2 replications in 2004. F<sub>4</sub> seed was increased by randomly selecting plants within each F<sub>3</sub> family. In 2005, a naturally infested field experiment was conducted with three replications of 38 resistant and 22 susceptible F<sub>4</sub> families. In both years plots were evaluated for powdery mildew severity using 1 through 5 scale (1 = no powder mildew – 5 = complete coverage of powdery mildew).

### **Results and Discussion**

The resistant and susceptible selections were not significantly different for mean powdery mildew severity in 2004 and in 2005 (Table 6). The resistant selection group may have more genetic variation, as this group had the most resistant (lowest minimum severity) and susceptible (highest maximum severity) genotypes in both years (Table 6). These results indicate that greenhouse selection of F<sub>2</sub> plants for powdery mildew resistance was not effective. Field selection is likely the preferable method to develop resistant germplasm.

### **Bolting Resistance**

High temperatures during fall plantings in the Yuma production region can result in premature bolting. Development of cultivars adapted to high temperature conditions are necessary to expand the production season in Yuma. Our breeding objective is to identify and select iceberg breeding lines with resistance to bolting and appropriate head characteristics for fall planting in Yuma, AZ.

### **Procedure**

Two field experiments were conducted in a single commercial field to evaluate yield and horticultural characteristics on 9 F<sub>6</sub> 87-714-8 x Autumn Gold. The first experiment was planted on September 15, 2005 with the check cultivars Autumn Gold, Lighthouse, Sahara, Salinas, Tiber, and Welton. The second experiment was planted on September 17, 2005 with the check cultivars Autumn Gold, Beacon, Heatmaster, Lighthouse, Prestige, Salinas, and Tiber. Each plot was evaluated at market maturity for head diameter, core length, head firmness, and rib protrusion. The percentage of harvested heads was determined and the cartons per acre for each plot was calculated from this value.

### **Results**

At the first planting date, significant differences were detected for head diameter and core length (Table 7). In general, the USDA breeding lines had larger heads with taller cores compared to the commercial cultivars. For the second planting date, significant differences were detected for head weight, head diameter, core length and rib protrusion (Table 7). As with the previous planting date, the USDA breeding lines had larger heads, taller cores, but also ribs that were more protruding compared to the cultivars. These lines have horticultural and yield characteristics that are distinct from current cultivars available for fall planting slots, despite having some negative characteristics such as tall cores and rib protrusion. Seed will be increased for further grower trials.

## Tipburn Resistance

Lettuce planted for harvest in March and April is exposed to high temperatures and increasing day length near market maturity, making the crop vulnerable to tipburn. Tipburn resistance is needed in cultivars adapted to late spring production to insure reliable quality for spring harvested lettuce. Additionally, romaine and leaf cultivars can be extremely susceptible to tipburn, and less breeding has occurred to improve leaf and romaine types for tipburn resistance. No reports exist on the tipburn resistance of leaf and romaine cultivars.

The objectives of this research are to:

- 1) Develop iceberg cultivars with a high level of tipburn resistance.
- 2) Determine the variation for tipburn resistance in romaine and leaf cultivars.
- 3) Identify romaine and leaf cultivars with tipburn resistance for use as parents in breeding.

## Procedure

A late spring tipburn trial was planned for planting on the Yuma Valley Agricultural Center to select iceberg breeding lines with superior tipburn resistance and to identify leaf and romaine cultivars with high tipburn resistance for use as parents in breeding. However, due to difficulties in scheduling water, this trial was not planted.

Late season plantings at the Yuma Agricultural Center works well for tipburn evaluations. However, they are not conducive for selection of head type due to high heat and advanced stages of plant deterioration. Therefore, a second field experiment was planted on January 2 near Texas Hill, AZ to evaluate horticultural characteristics, tipburn resistance, and estimate yield potential. Fifty foot plots were established for 21 F<sub>5</sub> advanced breeding lines of Salinas x Vanguard 75 and the check cultivars Gabilan, Navajo, Diamond, Head Master, Dominquez 67, and Green Lightning. The field cultivar was Gabilan. Five heads per plot were selected for collection of data on head weight (g), diameter (cm), core height (cm), rib protrusion (measured in ½ inch increments of protrusion), firmness (1 = no head; 2 = puffy head; 3 = firm head, yields under firm pressure; 4 = hard head, does not yield under pressure; 5 = splitting), and shape (round or pointed). The height of each head was also measured and used to calculate the volume of each head using the equation to estimate the volume of a sphere ( $\frac{4}{3}\pi \times \text{height} \times \frac{1}{2} \text{diameter}$ ). The density of each head was estimated by dividing head weight by head volume. Yield was estimated by determining the proportion harvestable heads for each plot after commercial harvest.

## Results and Discussion

Minimal tipburn was observed in the Texas Hill experiment, and was likely due to the unusually cool weather observed during this time period. Data were collected on yield and horticultural characters on April 1, 2006. The field was not at market maturity on this date, as evident from a mean firmness of 1 for the cultivar Gabilan (Table 8). Despite this, significant variation was observed for variables, and some breeding lines and cultivars performed acceptably at this earlier date. In particular, Navajo and the breeding line RH05-1361 had the best combination of firmness (between 3.0 – 4.0), head weight (> 700 g), diameter (> 15 cm), volume (>3000 cm<sup>3</sup>)

and density ( $>0.20 \text{ g/cm}^3$ ), with short cores ( $< 2 \text{ cm}$ ) and smooth ribs ( $< 2$ ). Given the unusual weather conditions, these results need to be confirmed with a second year's data.

## **Lettuce Drop (*Sclerotinia sclerotiorum*) resistance (Collaborative with B. Pryor)**

Lettuce drop is a destructive disease of lettuce caused by *Sclerotinia sclerotiorum* and *Sclerotinia minor* that occurs worldwide. Breeding for *S. minor* resistance in the Salinas Valley of California has been ongoing, and has identified sources of resistance in cultivated lettuce and developed breeding lines. *S. minor*, which predominates in the Salinas Valley, differs from *S. sclerotiorum* in that it infects lettuce only through eruptive germination. *S. sclerotiorum* predominates in the Yuma Valley, and can infect lettuce through eruptive germination of sclerotia or through airborne ascospores that infect lettuce heads. Breeding for resistance to *S. sclerotiorum* in Yuma has not previously been conducted. In 2004, a field experiment was conducted on the Yuma Valley Agricultural Research Center to determine if germplasm resistant to *S. minor* in the Salinas Valley is also resistant to *S. sclerotiorum* in Yuma, AZ. These results indicated that germplasm such as PI251246, Eruption, and Little Gem are resistant to both pathogens in both environments, while Batavia Reine des Glaces and Gladiator are susceptible. Additionally, iceberg breeding lines selected for resistance to *S. minor* in the Salinas Valley were resistant to *S. sclerotiorum* in Yuma, AZ.

The objectives of this research are to:

- 1) Determine if genotypes resistant to *S. minor* in the Salinas Valley are additionally resistant to *S. sclerotiorum* in Yuma, AZ.
- 2) Identify sources of resistance to *S. sclerotiorum*.

## **Procedure**

A field trial was conducted at the Yuma Valley Agricultural Center in collaboration with Dr. Barry Pryor (U of AZ). Sixty-one lines were planted on November 9, and include the *S. minor* susceptible checks Batavia Reine des Glaces and Gladiator, the resistant check PI251246, 30 *L. sativa* cultivars, 15 *L. sativa* breeding lines, 3 *L. virosa* accessions, and 6 BCF<sub>1:3</sub> *L. virosa* – *L. sativa* hybrid families. Sclerotia of *S. sclerotiorum* were incorporated into the beds prior to planting and each entry was then planted with 3 replications. Disease incidence was collected on March 28 and analyzed with Proc Mixed in SAS using arcsine transformed data.

## **Results and Discussion**

Variation for resistance was observed among the tested lines and families (Figure 1). The data from 2005 generally corresponded well with that from 2004, and the correlation between the two years was significant ( $r = 0.47$ ,  $P < 0.01$ ). Batavia Reine des Glaces and Gladiator was susceptible in both years. Most importantly, PI251246, Eruption, and Little Gem, performed well in both years, and indicate that these are useful lines for breeding resistance to lettuce drop caused by both pathogens in both environments. Some exceptions did occur; the leaf types



Tehema and Two Star were among the most susceptible in 2004, but were more resistant in 2005.

Three *L. virosa* accession were tested for resistance to lettuce drop in 2005 only, and were extremely resistant to the disease with less than 1% mortality. Variation for mortality was observed among BCF<sub>1:3</sub> hybrid families incorporating genes from accession IVT280, with disease incidence ranging from 11 – 32%. Selections were made from two families with disease incidence less than 17%.

The breeding lines selected for *S. minor* resistance in the Salinas Valley performed well in 2005. Six out of the most resistant 10 entries were iceberg breeding lines derived from Holborn Standard and Great Lakes 54. The data from 2004 and 2005 were pooled together used to calculate a contrast between 9 iceberg breeding lines, their parents (partially resistant Holborn Standard and Great Lakes 54, and susceptible Salinas), and the iceberg cultivars to determine the effect of selection for *S. minor* resistance on *S. sclerotiorum* resistance. These results indicated that the iceberg breeding lines were significantly more resistant from the iceberg cultivars in both years, but that the breeding lines and parents were not significantly different (Table 9). These results indicate that the breeding lines with previous selection for *S. minor* resistance are a better population for selection of *S. sclerotiorum* resistance than a randomly selected group of iceberg cultivars. Furthermore, this advantage appears to result from previous selection for resistance to *S. minor*, and from the use of parents with moderate levels of resistance to *S. minor*. Selections were made from the best performing breeding lines.

## **Fusarium root rot resistance (a.k.a Fusarium wilt)**

### **Procedure, Results, and Discussion:**

Fusarium wilt is a potential threat in California and is spreading in Arizona. Three physiological races of Fusarium are known in Japan. Our greenhouse and field data indicate that race 1 is present in California and Arizona. Limited greenhouse data from F<sub>1</sub> individuals and three F<sub>2</sub> families indicate that resistance to race 1 in ‘Costa Rica No.4’ and ‘Salinas’ is controlled by recessive genes.

A naturally-infected field test in Yuma (planted September 20, 2005) was not satisfactory for selection of resistant F<sub>2</sub> segregants as nearly 50% of the plants were asymptomatic for wilt (Figure 2). The greenhouse F<sub>2</sub> data would have predicted 321 susceptible: 107 resistant or 401 susceptible: 27 resistant. Cultivar rankings in this field test were consistent with previous data (Table 10). ‘Appollo’, BOS 9021, ‘Conquistador’, ‘Fresheart’, ‘Slugger’, and ‘King Louie’ appeared more resistant than ‘Salinas’. Data from eight F<sub>3</sub> families clearly showed the high frequency of escapes in F<sub>2</sub> families (Table 11), and underscore the importance of uniform, high level of disease expression in selection for resistance to Fusarium wilt.

Table 1. Percent big vein symptomatic plants among 17 cultivars tested in an infested Yuma, AZ field experiment.

| Line          | Type  | Percent symptomatic | 95 % confidence interval |       |
|---------------|-------|---------------------|--------------------------|-------|
|               |       |                     | Lower                    | Upper |
| Pavane        | Latin | 3.3                 | 0.3                      | 26.3  |
| Wintersselect | Head  | 17.0                | 5.0                      | 44.2  |
| Pacific       | Head  | 25.7                | 9.3                      | 54.0  |
| Coyote        | Head  | 28.9                | 11.1                     | 56.8  |
| Honcho        | Head  | 29.3                | 11.4                     | 57.2  |
| Winterhaven   | Head  | 36.7                | 15.2                     | 65.2  |
| Red Coach 74  | Head  | 48.5                | 24.0                     | 73.8  |
| Grizzly       | Head  | 51.1                | 25.9                     | 75.7  |
| Valley Queen  | Head  | 51.3                | 26.1                     | 75.9  |
| Badger        | Head  | 51.6                | 26.3                     | 76.1  |
| Yuma          | Head  | 54.2                | 28.4                     | 78.0  |
| Red Coach     | Head  | 56.3                | 30.1                     | 79.4  |
| Del Oro       | Head  | 57.0                | 30.7                     | 79.9  |
| Winter King   | Head  | 59.2                | 32.5                     | 81.3  |
| Vanguard 75   | Head  | 62.8                | 35.7                     | 83.7  |
| Desert Storm  | Head  | 63.1                | 36.0                     | 83.9  |
| Cibola        | Head  | 68.7                | 41.3                     | 87.3  |

Table 2. Big vein resistance in Pacific x (Pacific x Pavane) breeding line in 2004 and 2005 Yuma, AZ field experiments.

| Line          | Pedigree                     | Number tested | Percent symptomatic |
|---------------|------------------------------|---------------|---------------------|
| <i>2004</i>   |                              |               |                     |
| Pavane        | Latin                        | 100           | 6.0                 |
| 01-2060       | Pacific x (Pacific x Pavane) | 98            | 10.2                |
| 01-2058       | Pacific x (Pacific x Pavane) | 107           | 10.3                |
| 01-2059       | Pacific x (Pacific x Pavane) | 106           | 17.0                |
| 01-2062       | Pacific x (Pacific x Pavane) | 106           | 22.6                |
| 01-2063       | Pacific x (Pacific x Pavane) | 96            | 22.9                |
| Pacific       | iceberg                      | 109           | 22.9                |
| Wintersselect | iceberg                      | 364           | 40.7                |
| Winterhaven   | iceberg                      | 65            | 75.4                |
| Vanguard 75   | iceberg                      | 67            | 82.1                |
| <i>2005</i>   |                              |               |                     |
| Line          |                              |               |                     |
| Pavane        | Latin                        | 166           | 3.0                 |
| RH05-0707     | Pacific x (Pacific x Pavane) | 126           | 10.3                |
| RH05-0696     | Pacific x (Pacific x Pavane) | 117           | 11.1                |
| RH05-0692     | Pacific x (Pacific x Pavane) | 125           | 13.6                |
| Wintersselect | Iceberg                      | 122           | 15.6                |
| RH05-0708     | Pacific x (Pacific x Pavane) | 131           | 16.8                |
| RH05-0691     | Pacific x (Pacific x Pavane) | 127           | 17.3                |
| RH05-0695     | Pacific x (Pacific x Pavane) | 126           | 17.5                |
| RH05-0699     | Pacific x (Pacific x Pavane) | 126           | 19.0                |
| RH05-0693     | Pacific x (Pacific x Pavane) | 127           | 19.7                |
| RH05-0706     | Pacific x (Pacific x Pavane) | 125           | 20.8                |
| RH05-0703     | Pacific x (Pacific x Pavane) | 127           | 21.3                |
| RH05-0690     | Pacific x (Pacific x Pavane) | 129           | 21.7                |
| RH05-0694     | Pacific x (Pacific x Pavane) | 110           | 21.8                |
| RH05-0702     | Pacific x (Pacific x Pavane) | 128           | 22.7                |
| RH05-0700     | Pacific x (Pacific x Pavane) | 121           | 23.1                |
| Pacific       | Iceberg                      | 175           | 24.0                |
| RH05-0701     | Pacific x (Pacific x Pavane) | 121           | 28.9                |
| RH05-0697     | Pacific x (Pacific x Pavane) | 124           | 29.0                |
| RH05-0698     | Pacific x (Pacific x Pavane) | 118           | 30.5                |
| RH05-0705     | Pacific x (Pacific x Pavane) | 127           | 31.5                |
| Winterhaven   | Iceberg                      | 165           | 35.2                |
| RH05-0704     | Pacific x (Pacific x Pavane) | 122           | 39.3                |
| Vanguard 75   | Iceberg                      | 127           | 60.6                |

Table 3. Big vein disease area under the disease progress curve (AUDPC) for *L. virosa* accession IVT280, BCF<sub>3</sub> virosa-sativa hybrids 00-366-3 and 00-366-9 and 5 cultivars.

| Genotype       | Number of<br>Plants Tested | Mean AUDPC <sup>1</sup> | Percent<br>Symptomatic |
|----------------|----------------------------|-------------------------|------------------------|
| IVT280         | 65                         | 0.0 a                   | 0                      |
| Pacific        | 68                         | 5.6 ab                  | 32                     |
| Pavane         | 66                         | 6.7 ab                  | 56                     |
| 00-366-3       | 69                         | 8.2 ab                  | 41                     |
| Clemente       | 68                         | 10.3 bc                 | 53                     |
| 00-366-9       | 69                         | 13.7 bc                 | 45                     |
| Salinas 88     | 70                         | 19.2 d                  | 86                     |
| Great Lakes 65 | 71                         | 22.6 d                  | 73                     |

1/ means followed by different letters are significantly different (p < 0.05)

Table 4. Distribution of big vein area under the disease progress curve (AUDPC) in BC<sub>2</sub>F<sub>4</sub> *L. virosa* - *L. sativa* hybrid families.

| Pedigree              | Number<br>of families<br>tested | AUDPC |            | Percent more<br>resistant <sup>1</sup> than<br><i>L. sativa</i> parent | Percent more<br>resistant <sup>1</sup> than<br>both parents |
|-----------------------|---------------------------------|-------|------------|--|---|
|                       |                                 | Mean  | Min - Max  |  |   |
| 03-366-9 x Salinas    | 8                               | 12.3  | 2.8 – 25.2 | 40%  | 7%  |
| 03-366-9 x Salinas 88 | 7                               | 11.3  | 2.3 – 21.3 | 25%  | 5%  |
| 03-366-3 x Salinas 88 | 4                               | 7.8   | 1.9 – 16.8 | 58%  | 0%  |
| 03-366-3 x Salinas    | 7                               | 13.8  | 3.8 – 31.8 | 20%  | 0%  |
| 03-366-3 x Pacific    | 9                               | 10.7  | 1.6 – 30.3 | 0%   | 0%  |
| 03-366-9 x Clemente   | 15                              | 8.3   | 0.3 – 22.5 | 0%   | 0%  |
| 03-366-3 x Pavane     | 14                              | 7.6   | 0.8 – 23.4 | 0%   | 0%  |

1/ Families significantly better at p < 0.05

Table 5. Big vein area under the disease progress curve and percent symptomatic plants of 6 plant introductions, Pavane, Pacific, Margarita, and Great Lakes 65 in a greenhouse experiment.

| Line           | Number plants tested | Percent symptomatic | Mean  | AUDPC                   |       |
|----------------|----------------------|---------------------|-------|-------------------------|-------|
|                |                      |                     |       | 95% confidence interval |       |
|                |                      |                     |       | Lower                   | Upper |
| Pavane         | 35                   | 2.9                 | 0.11  | 0.00                    | 6.19  |
| Margarita      | 36                   | 11.1                | 2.14  | 0.00                    | 8.23  |
| PI342452c      | 35                   | 31.4                | 3.76  | 0.00                    | 9.84  |
| PI342555       | 36                   | 41.7                | 4.57  | 0.00                    | 10.66 |
| PI324242       | 36                   | 36.1                | 7.88  | 1.79                    | 13.96 |
| PI342462       | 36                   | 55.6                | 7.88  | 1.79                    | 13.96 |
| Pacific        | 36                   | 41.7                | 8.07  | 1.98                    | 14.16 |
| PI342510       | 35                   | 51.4                | 10.95 | 4.86                    | 17.04 |
| PI320467       | 34                   | 44.1                | 11.48 | 5.39                    | 17.57 |
| Great Lakes 65 | 36                   | 83.3                | 26.44 | 20.36                   | 32.53 |

Table 6. Mean and genotype range of powdery mildew severity in naturally infested field experiment near Yuma, AZ in 2004 and 2005.

| Selection group        | Mean severity | 95 % confidence interval |       | Range of genotype means |
|------------------------|---------------|--------------------------|-------|-------------------------|
|                        |               | Lower                    | Upper |                         |
| 2004                   |               |                          |       |                         |
| Soraya                 | 2.8           | 2.2                      | 3.3   | NA                      |
| Salinas                | 4.9           | 4.4                      | 5.4   | NA                      |
| Resistant Selections   | 2.3           | 2.1                      | 2.6   | 1.9 - 3.7               |
| Susceptible Selections | 2.4           | 1.8                      | 3.1   | 2.0 - 3.1               |
| 2005                   |               |                          |       |                         |
| Soraya                 | 2.3           | 1.3                      | 3.3   | NA                      |
| Salinas                | 2.9           | 2.2                      | 3.6   | NA                      |
| Resistant Selections   | 2.3           | 1.6                      | 3.9   | 1.4 - 3.6               |
| Susceptible Selections | 2.5           | 1.8                      | 3.2   | 1.8 - 3.2               |

Table 7. Horticultural characteristics of 87-714-8 x Autumn Gold F6 breeding lines, parents, and check cultivars two Yuma, AZ field trials

| Line                        | Head<br>Wieght       | Head<br>Diameter | Core<br>Length | Head<br>Firmness   | Rib<br>Protrusion  | Cartons<br>per acre | Harvest<br>Percentage |
|-----------------------------|----------------------|------------------|----------------|--------------------|--------------------|---------------------|-----------------------|
| <i>September 15 Harvest</i> |                      |                  |                |                    |                    |                     |                       |
| Autumn Gold                 | 1310.0 <sup>ns</sup> | 14.0 abc         | 2.9 ab         | 2.33 <sup>ns</sup> | 1.38 <sup>ns</sup> | 979 <sup>ns</sup>   | 0.90 ab               |
| Lighthouse                  | 1541.7               | 12.9 a           | 3.5 ab         | 3.6                | 1.1                | 881                 | 0.82 a                |
| Sahara                      | 1332.0               | 13.2 ab          | 1.3 a          | 1.9                | 1.8                | 997                 | 0.92 ab               |
| Salinas                     | 1504.2               | 14.8 abcd        | 4.9 b          | 2.1                | 1.9                | 948                 | 0.88 ab               |
| Tiber                       | 1506.3               | 16.3 cd          | 4.5 b          | 2.5                | 1.6                | 1004                | 0.93 b                |
| Wellton                     | 1637.0               | 15.3 abcd        | 3.2 ab         | 3.5                | 0.8                | 1013                | 0.93 ab               |
| RH04-1103-2                 | 1567.4               | 15.3 abcd        | 5.3 b          | 2.4                | 2.6                | 1018                | 0.94 ab               |
| RH04-1103-4                 | 1599.8               | 17.0 cd          | 4.1 ab         | 2.8                | 1.6                | 1008                | 0.93 ab               |
| RH04-1103-5                 | 1640.6               | 16.9 d           | 5.0 b          | 2.7                | 1.8                | 1035                | 0.96 b                |
| RH04-1103-6                 | 1533.0               | 15.1 abcd        | 3.9 ab         | 3.2                | 2.2                | 986                 | 0.91 ab               |
| RH04-1104-1                 | 1459.8               | 17.1 d           | 3.8 ab         | 2.4                | 1.5                | 1062                | 0.98 ab               |
| RH04-1104-4                 | 1603.0               | 16.4 bcd         | 4.4 ab         | 2.8                | 2.6                | 823                 | 0.76 a                |
| <i>September 17 Harvest</i> |                      |                  |                |                    |                    |                     |                       |
| Autumn Gold                 | 1246.4 a             | 13.2 ab          | 1.9 a          | 3.00 <sup>ns</sup> | 2.1 abc            | 1035 <sup>ns</sup>  | 0.96 <sup>ns</sup>    |
| Beacon                      | 1484.2ab             | 15.4 ab          | 1.8 a          | 2.7                | 1.4 ab             | 1056                | 0.98                  |
| Heatmaster                  | 1466.6 ab            | 13.4 ab          | 3.0 ab         | 3.5                | 1.7 ab             | 1029                | 0.95                  |
| Lighthouse                  | 1659.2 b             | 12.9 a           | 2.4 ab         | 4.0                | 0.6 a              | 1040                | 0.96                  |
| Prestige                    | 1574.0 ab            | 13.2 ab          | 2.9 ab         | 3.8                | 1.4 ab             | 948                 | 0.88                  |
| Salinas                     | 1474.6 ab            | 15.7 ab          | 3.5 ab         | 2.6                | 2.0 ab             | 980                 | 0.91                  |
| Tiber                       | 1648.4 b             | 16.7 ab          | 4.2 b          | 3.0                | 2.3 abc            | 1008                | 0.93                  |
| RH04-1103-1                 | 1847.3 b             | 17.6 a           | 4.3 b          | 3.1                | 3.0 bc             | 1040                | 0.96                  |
| RH04-1103-2                 | 1507.7 ab            | 15.4 ab          | 2.8 ab         | 2.7                | 3.3 bc             | 1051                | 0.97                  |
| RH04-1103-3                 | 1578.1 ab            | 15.2 ab          | 2.9 ab         | 3.1                | 2.6 abc            | 1008                | 0.93                  |
| RH04-1103-4                 | 1817.7 b             | 16.8 ab          | 4.3 b          | 3.5                | 4.2 c              | 986                 | 0.91                  |
| RH04-1103-5                 | 1693.8 b             | 16.7 ab          | 3.4 ab         | 2.9                | 3.0 bc             | 1024                | 0.95                  |
| RH04-1103-6                 | 1645.1 ab            | 17.1 ab          | 4.2 b          | 3.1                | 2.9 bc             | 1051                | 0.97                  |
| RH04-1104-1                 | 1450.1 ab            | 15.8 ab          | 2.6 ab         | 2.9                | 2.8 bc             | 1040                | 0.96                  |
| RH04-1104-4                 | 1705.3 b             | 15.1 ab          | 4.2 b          | 3.7                | 2.7 abc            | 1029                | 0.95                  |
| RH04-1104-6                 | 1678.5 ab            | 16.7 ab          | 3.4 ab         | 2.9                | 3.5 bc             | 1062                | 0.98                  |

Numbers followed by different letters are significantly different at  $P < 0.05$ , ns = no significantly different.



Table 8. Horticultural characteristics of 6 cultivars and 21 Salinas x Vanguard 75 F<sub>4,5</sub> breeding lines in 2005 Texas Hill, AZ field experiment.

| Line             | Weight<br>(grams) | Diameter<br>(cm) | Volume<br>(cm <sup>3</sup> ) | Density<br>(g/cm <sup>3</sup> ) | Core height<br>cm | Rib<br>protrusion <sup>1</sup> | Firmness <sup>2</sup> | Shape        | Harvest<br>percentage |
|------------------|-------------------|------------------|------------------------------|---------------------------------|-------------------|--------------------------------|-----------------------|--------------|-----------------------|
| Diamond          | 577 abcd          | 16.4 c           | 4069 cdef                    | 0.142 a                         | 2.8 bcde          | 1.7 acd                        | 2.0 abcd              | round        | 98                    |
| Dominguez 67     | 537 abc           | 15.1 abc         | 3343 abcdef                  | 0.167 ab                        | 2.5 abcde         | 1.0 a                          | 2.6 bcde              | round        |                       |
| Gabilan          | 494 ab            | 13.1 a           | 2524 ab                      | 0.208 abc                       | 1.6 a             | 1.5 ac                         | 1.0 a                 | round        | 98                    |
| Green Lightning  | 475 a             | 13.2 ab          | 2431 ab                      | 0.204 abc                       | 2.1 abcde         | 1.2 ac                         | 3.0 cde               | round        |                       |
| Head Master      | 566 abcd          | 15.2 abc         | 3501 abcdef                  | 0.162 ab                        | 2.3 abcde         | 1.2 ac                         | 2.8 cde               | round        |                       |
| <b>Navajo</b>    | <b>740 cdef</b>   | <b>15.8 abc</b>  | <b>3701 abcdef</b>           | <b>0.203 abc</b>                | <b>2.0 abcd</b>   | <b>1.5 ac</b>                  | <b>3.6 e</b>          | <b>round</b> | <b>100</b>            |
| <b>RH05-1361</b> | <b>736 cdef</b>   | <b>15.0 abc</b>  | <b>3416 abcdef</b>           | <b>0.232 abc</b>                | <b>2.3 abcde</b>  | <b>1.2 ac</b>                  | <b>3.0 cde</b>        | <b>round</b> | <b>96</b>             |
| RH05-1362        | 544 abcd          | 15.7 abc         | 3695 abcdef                  | 0.154 a                         | 2.6 abcde         | 2.7 acde                       | 2.4 abcde             | round        | 98                    |
| RH05-1363        | 516 abc           | 14.3 abc         | 3433 abcdef                  | 0.167 ab                        | 2.6 abcde         | 4.4 de                         | 2.2 abcde             | round        | 99                    |
| RH05-1364        | 596 abcd          | 14.2 abc         | 3236 abcdef                  | 0.219 abc                       | 3.1 de            | 1.2 ac                         | 2.6 bcde              | round        | 97                    |
| RH05-1365        | 690 abcd          | 16.3 bc          | 4505 f                       | 0.159 a                         | 2.9 cde           | 1.5 ac                         | 2.8 cde               | round        | 97                    |
| RH05-1366        | 743 cdef          | 16.5 c           | 4170 def                     | 0.181 ab                        | 2.8 bcde          | 1.7 acd                        | 3.0 cde               | round        | 97                    |
| RH05-1367        | 580 abcd          | 14.5 abc         | 3220 abcdef                  | 0.187 ab                        | 2.5 abcde         | 2.0 acd                        | 3.6 e                 | round        | 97                    |
| RH05-1368        | 646 abcd          | 15.7 abc         | 3874 bcdef                   | 0.169 ab                        | 3.2 e             | 1.7 acd                        | 3.4 de                | round        | 100                   |
| RH05-1369        | 799 ef            | 14.1 abc         | 2995 abcde                   | 0.280 bc                        | 2.1 abcde         | 2.0 acd                        | 2.8 cde               | round        | 99                    |
| RH05-1370        | 546 abcd          | 13.2 ab          | 2579 abc                     | 0.220 abc                       | 2.5 abcde         | 1.0 a                          | 2.6 bcde              | round        | 96                    |
| RH05-1372        | 594 abcd          | 15.2 abc         | 3511 abcdef                  | 0.176 ab                        | 3.1 de            | 3.2 acde                       | 1.6 abc               | round        | 92                    |
| RH05-1373        | 703 abcd          | 13.2 ab          | 2738 abcd                    | 0.263 bc                        | 1.7 ab            | 3.0 acde                       | 3.2 de                | pointed      | 95                    |
| RH05-1374        | 721 bcdef         | 15.0 abc         | 3206 abcdef                  | 0.243 abc                       | 1.8 abc           | 3.4 acde                       | 2.2 abcde             | round        | 93                    |
| RH05-1377        | 788 ef            | 13.0 a           | 2665 abcd                    | 0.300 c                         | 1.7 a             | 4.0 cde                        | 3.4 de                | pointed      | 97                    |
| RH05-1378        | 663 abcd          | 14.6 abc         | 3298 abcdef                  | 0.224 abc                       | 2.6 abcde         | 4.9 e                          | 1.8 abc               | round        | 95                    |
| RH05-1380        | 871 f             | 13.8 abc         | 2930 abcd                    | 0.280 bc                        | 2.5 abcde         | 2.2 acde                       | 2.8 cde               | round        | 98                    |
| RH05-1381        | 777 def           | 16.9 c           | 4498 ef                      | 0.182 ab                        | 2.7 abcde         | 2.0 acd                        | 3.6 e                 | round        | 99                    |

<sup>1</sup> rib protrusion: estimated in 1/2 in increments of the most protruding rib.

<sup>2</sup> firmness: 1 = no head; 2 = puffy head; 3 = firm head (yields under firm pressure); 4 = hard head (does not yield under pressure); 5 = splitting head.

Numbers followed by the same letter are not significantly different at P < 0.05.



Table 9. Contrast between 9 iceberg breeding lines, their parents, and modern iceberg cultivars for lettuce drop disease incidence due to *Sclerotinia sclerotiorum* in 2004 and 2005 inoculated field experiments in Yuma, AZ.

| Group                  | 2004 Disease Rating |      |                                      | 2005 Disease Rating |      |                         |
|------------------------|---------------------|------|--------------------------------------|---------------------|------|-------------------------|
|                        | No of lines         | Mean | Contrast <sup>1</sup><br>F statistic | No of lines         | Mean | Contrast<br>F statistic |
| Iceberg Breeding Lines | 9                   | 0.29 |                                      | 8                   | 0.16 |                         |
| Parents <sup>2</sup>   | 3                   | 0.42 | 3.72                                 | 3                   | 0.26 | 0.96                    |
| Iceberg Cultivars      | 8                   | 0.51 | 18.16**                              | 10                  | 0.29 | 16.32**                 |

1/ All contrast are versus iceberg breeding lines

2/ Holborn Standard, Great Lakes 65, and Salinas

Table 10. Percent lettuce plants exhibiting Fusarium wilt symptoms in fall lettuce, Yuma.

| Entry           | Type <sup>z</sup> | % infected | Entry       | Type <sup>z</sup> | % infected |
|-----------------|-------------------|------------|-------------|-------------------|------------|
| Costa Rica #4   | IR                | 0          | Apollo      | R                 | 10.5       |
| Banchu Red Fire | L                 | 0          | Salinas     | I                 | 21.3       |
| BOS 9021        | R                 | 0          | Merit       | I                 | 21.7       |
| Conquistador    | R                 | 0          | BOS 9032    | R                 | 38.5       |
| Fresheart       | R                 | 0          | Empire      | I                 | 43.5       |
| Slugger         | R                 | 2          | BOS 9034    | R                 | 51.7       |
| King Louie      | R                 | 3.2        | Autumn Gold | I                 | 57.4       |
| Calmar          | I                 | 3.6        | Patriot     | IR                | 83.3       |
| Salinas 88      | I                 | 4          | Climax      | I                 | 83.9       |
| Rivergreen      | IR                | 6.9        | 8830        | I                 | 88.9       |
| Grand Rapids    | L                 | 9.1        | Vanguard    | I                 | 92.7       |

<sup>z</sup>I, iceberg; R, romaine; IR, iceberg-romaine; L, leaf.

Table 11. Fusarium wilt symptom rating of eight F<sub>2</sub> segregants, their respective F<sub>3</sub> family Fusarium wilt symptom data, and the F<sub>2</sub> genotype for Fusarium wilt; ‘Salinas 88’ and ‘Vanguard’ for comparison; greenhouse tests.

| F <sub>2</sub> plant rating | F <sub>3</sub> family data |   |   |   | F <sub>2</sub> genotype |
|-----------------------------|----------------------------|---|---|---|-------------------------|
|                             | 1                          | 2 | 3 | 4 |                         |
| 1                           | 0                          | 0 | 2 | 8 | S                       |
| 2                           | 4                          | 3 | 2 | 1 | r                       |
| 1                           | 0                          | 1 | 4 | 5 | S                       |
| 1                           | 1                          | 3 | 2 | 4 | S                       |
| 1                           | 5                          | 1 | 2 | 2 | r                       |
| 1                           | 2                          | 3 | 1 | 4 | S                       |
| 1                           | 1                          | 3 | 3 | 3 | S                       |
| 1                           | 8                          | 1 | 0 | 1 | r                       |
| Salinas 88                  | 8                          | 2 | 0 | 0 |                         |
| Vanguard                    | 2                          | 0 | 4 | 4 |                         |

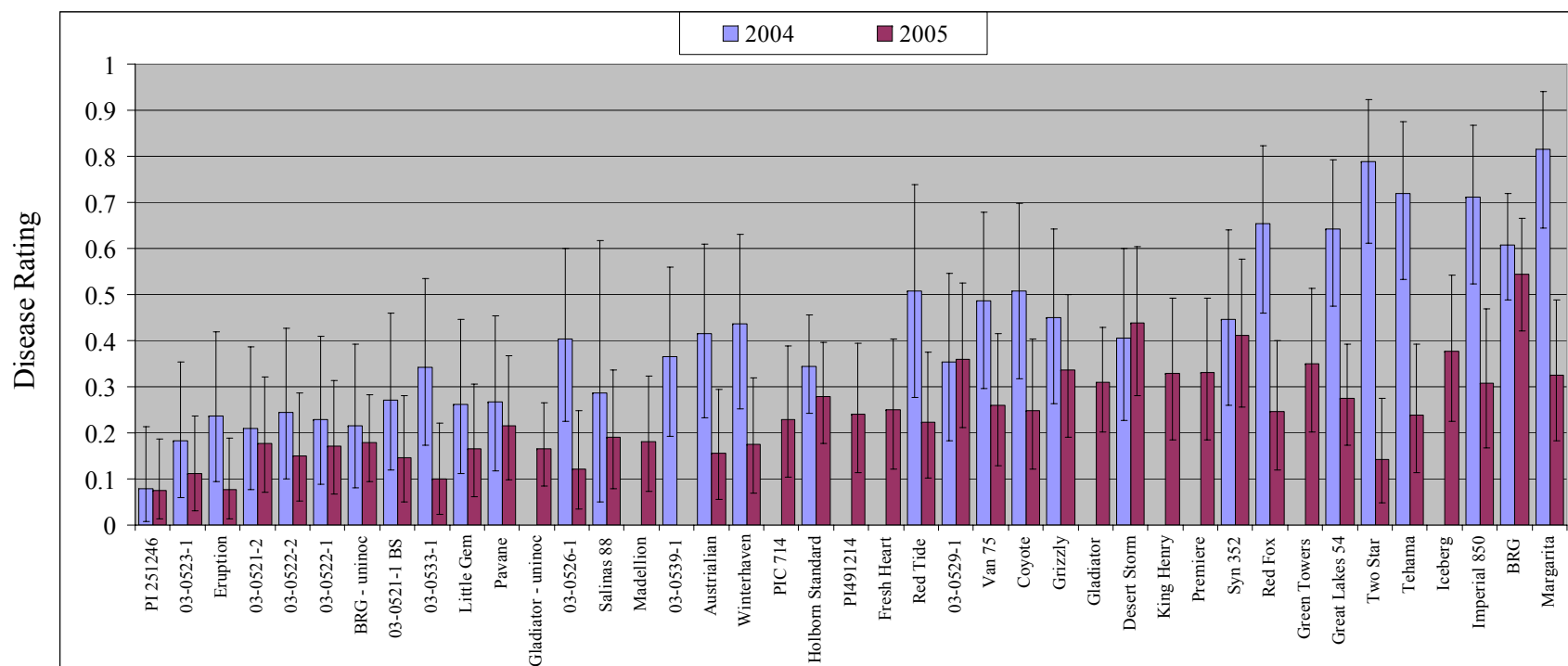


Figure 1. Variation in lettuce drop mortality among *Lactuca* germplasm in 2004 and 2005 *Sclerotinia sclerotiorum* infested field experiments in Yuma, AZ. BRG is Batavia Riene des Glaces. Uninoc is uninoculated controls. Error bars indicate 95% confidence intervals. Disease rating is derived from an arcsine transformation of the disease incidence.

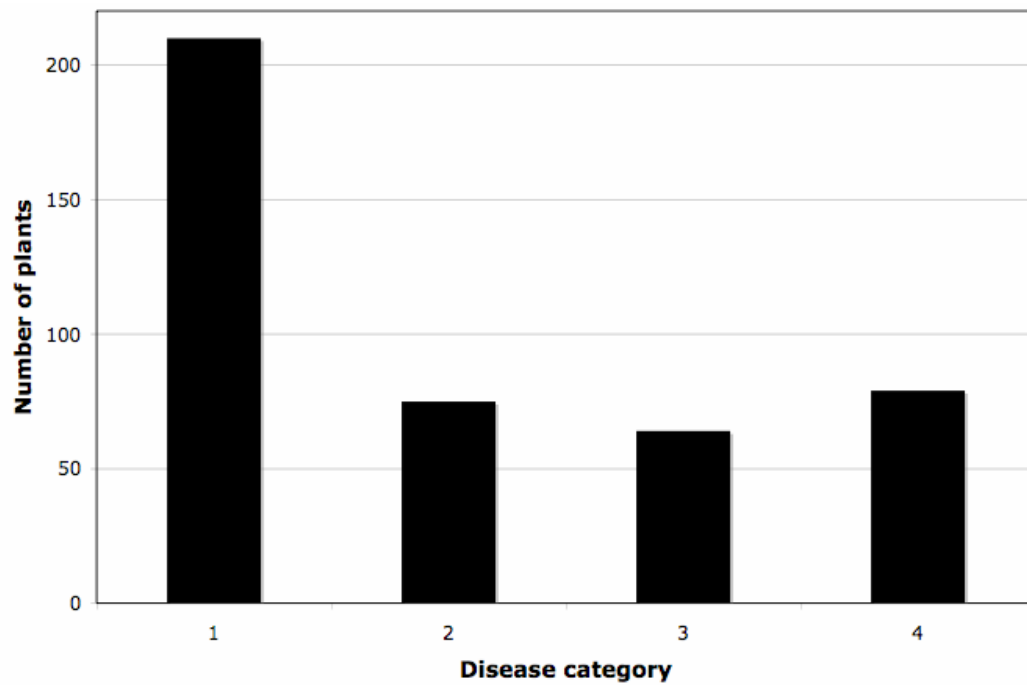


Figure 2. Numbers of  $F_2$  segregants by Fusarium wilt disease category (1, asymptomatic; 2, slight-moderate stunting; 3, severe stunting and yellowing; 4, senesced), Yuma field test.